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BENTHOCTOPUS RIGBYAE, N. SP., A NEW SPECIES OF CEPHALOPOD (OCTOPODA; INCIRRATA) FROM NEAR THE ANTARCTIC PENINSULA

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ABSTRACT

Among the many octopods collected during recent Antarctic trawling surveys were 93 specimens of an undescribed octopod with biserial suckers. Although most similar in appearance to the sub-Antarctic *Benthoctopus levis* (Hoyle 1885), these octopods differed in several morphological characters, including arm length, web depth, and details of the hectocotylus. Furthermore, molecular evidence supports the separation of the present material from known species. We therefore describe a new species named ***Benthoctopus rigbyae****.* These octopods attain mantle lengths of at least 105 mm (400 mm total length), and they are common at depths of 250–600 m throughout the South Shetland Island chain off the Antarctic Peninsula. We present some information on the biology of this species.
Keywords: deep-sea, Southern Ocean, Octopodidae, Antarctica.

INTRODUCTION

Incirrate octopods are common members of the Antarctic marine benthos (Dell, 1972; Allcock et al., 2001). Most of the octopods collected in Antarctic samples belong to genera with uniserial suckers, such as *Pareledone, Megaleledone,* and their relatives (e.g., Allcock, 2005; Daly & Rodhouse, 1994; Kuehl, 1988; Kubodera & Okutani, 1994; Lu & Stranks, 1994; Taki, 1961). During recent trawling cruises in the vicinity of the Antarctic Peninsula (Piatkowski et al., 1998, 2003; Barratt & Jorgensen, 2008), we collected 93 specimens of an octopod possessing biserial suckers, but lacking an ink sac. These octopods, which came from depths of about 200–600 m, are similar to *Benthoctopus levis* (Hoyle, 1885). The type locality of *B. levis* is at a depth of about 140 m, near Heard Island, a sub-Antarctic Island in the Indian Ocean. The peninsula specimens differ from *B. levis* in relative arm length, web depth, and hectocotylus details. We therefore consider the peninsula specimens to be members of a newly discovered species of *Benthoctopus*, which we here name ***B. rigbyae****.*

MATERIALS AND METHODS

R/V *Polarstern*'s cruises ANT XIV/2 (November–December 1996), ANT XIX/3 (January–February 2002), and ANT XXIII/8 (December 2006–January 2007) focused on the South Shetland Islands northwest of the Antarctic Peninsula. One research component of each cruise was a fisheries survey using commercial-sized bottom trawls (Piatkowski et al., 1998, 2003). Sampling was conducted on ANT XIV/2 at 40 stations around Elephant Island based on a stratified-random survey design in depths of 100–500 m. Two transects of stations northwest of King George Island at depths of 400, 600, and 800 m were also sampled with the same gear. A similar survey during ANT XIX/3 trawled at 49 stations around Elephant Island, 21 stations from 100–500 m depth in the southern South Shetlands, and five stations on the shelf north of Joinville Island. ANT XXIII/8 sampled 51 bottom-trawl stations around Elephant Island and another 38 stations across the Bransfield Straits close to the Peninsula, around Joinville Island and in the western Weddell Sea. Additionally, 23 samples were

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TABLE 1. Criteria used for male and female maturity stages.

	Male	Female
Stage 1	Sex undeterminable	Sex undeterminable
Stage 2	Testes developing or developed, but no spermatophores developing	Ovary small and round, eggs tiny
Stage 3	Spermatophores developing but none are mature	Eggs begin to enlarge
Stage 4	Few mature spermatophores present in Needham's sac	Ovaries swollen and oviducal glands enlarged
Stage 5	Four or more mature spermatophores present in Needham's sac and terminal organ	Ovaries completely distended with many mature eggs
Stage 6	Needham's sac swollen but empty	Ovaries swollen but empty, oviducal glands enlarged

collected with an Agassiz beam-trawl on ANT XIV/2, nine with a smaller Agassiz trawl on ANT XIX/3, and 15 on ANT XXIII/8, at various depths between 100–5,200 m. Details of all stations, including those that did not yield *Benthoctopus*, can be found in the cruise reports (Arntz & Brey, 1997; Kattner et al., 1998; Barratt & Jorgensen, 2008). Details of stations yielding *Benthoctopus* are provided in Figure 8.

All cephalopods were retrieved from all samples. Station locations and distribution of these cephalopods are presented by Piatkowski et al. (1998, 2003) and Barratt & Jorgensen (2008). Dorsal mantle length (ML) and total length were measured and sex and maturity stage determined according to set criteria (Table 1) for all specimens.

Under material examined, we list stage 1 as juvenile, stages 2–3 as immature, stages 4–5 as mature, and stage 6 as spent. Additionally for 15 males and 12 females representing the full range of size and maturity encountered, the following measurements and counts (following Roper & Voss, 1983) were recorded prior to fixation: measurements (Tables 2, 3) – ventral mantle length, mantle width, head width, eye length, width of pallial aperture, full funnel length, free funnel length, web formula, depth of deepest web segment, arm width, length of each arm, sucker diameter; counts (Table 4) – numbers of suckers on left (males and females) and right (males only) ventrolateral arms, number of suckers on the longest arm, number of inner and outer gill lamellae. The lengths of the calamus and ligula were recorded for males and, for six mature males, the length and width of a mature spermatophore as well. For six mature females, the length and width of the largest ovarian egg was measured.

Some specimens were dissected to examine internal anatomy prior to fixation. Dissections emphasized male and female reproductive anatomy and comparative anatomy of digestive tracts. Eleven females and 24 males were weighed to examine the relationship between length and weight. Stomach contents were determined for a few specimens, selected arbitrarily.

Molecular Analyses

Tissue samples were taken from two *Benthoctopus* specimens from the Antarctic Peninsula (NMSZ 2002037.032 and USNM 1021054) and preserved in 70% ethanol. Tissue samples were also preserved in 70% ethanol from a specimen of *Benthoctopus* sp. captured in the eastern Weddell Sea and from two specimens of *Benthoctopus levis* captured around Heard Island. We also obtained a beak taken from a *Benthoctopus* specimen presumed to be *B. thielei* Robson, 1932, captured off Kerguelen Island, and extracted DNA from this. Although DNA has previously been extracted from beach-washed *Spirula* shells (Strugnell et al., 2006), we believe that this is the first published report of DNA from a cephalopod beak.

The DNA extraction protocol followed that given in Allcock et al. (2006). Primers for three mitochondrial genes (12S rDNA, 16S rDNA, COI) were taken from the literature (Simon et al., 1990, 1991; Folmer et al., 1994), with the 12S rDNA and COI primers modified slightly to match cephalopod sequences on GenBank. Polymerase chain reactions (PCR) were carried out in 25 ml volumes. Thermal cycling conditions consisted of a denaturation step at 94°C for 2 min, followed by 35 cycles of 94°C

TABLE 2. Measurements (mm) of type specimens of ***Benthoctopus rigbyae*** n. sp. as archived at NMNH. Dorsal mantle length (prior to fixation) was measured on freshly collected specimens onboard ship.

	Holotype	Paratypes	
Sex	male	female	female
Total length	229	259	243
Dorsal mantle length (prior to fixation)	64	55	52
Dorsal mantle length (preserved)	49	47	42
Ventral mantle length	43	46	38
Mantle width	37	45	33
Head width	23	28	25
Eye diameter	9	8	6
Pallial aperture width	25	29	26
Full funnel length	16	17	14
Free funnel length	12	13	10
Deepest web segment depth	26	47	30
Arm width (longest arm)	5	4	5
Left dorsal arm length	176	200	168
Left dorsolateral arm length	175	202	169
Left ventrolateral arm length	147	201	180
Left ventral arm length	164	196	166
Right dorsal arm length	179	200	165
Right dorsolateral arm length	156	182	179
Right ventrolateral arm length	127	189	167
Right ventral arm length	166	183	172
Diameter of largest sucker	5	6	5
Calamus length	6		
Ligula length	17		
Web depth, segment A	21	47	29
Web depth, segment B	26	33	28
Web depth, segment C	26	29	damaged
Web depth, segment D	25	29	30
Web depth, segment E	25	16	24

for 40 s, 50°C for 40 s, and 72°C for 90 s. A final extension step of 72°C for 10 min was added in each case. Annealing temperatures varied according to the primers used and are available from the authors on request. Amplified products were purified using the QiaGen PCR purification kit following the manufacturer's instructions. Purified PCR products were commercially sequenced by Macrogen Inc. in both directions using the same primers used for PCR amplification. Sequences for *Enteroctopus dofleini* (Wülker, 1910), *Benthoctopus normani* (Massy, 1907), *Benthoctopus eureka* (Robson, 1932), and *Benthoctopus johnsoniana* Allcock et al., 2006, collected from the northeastern Pacific, the Falkland Islands and the northeastern Atlantic respectively were available from previous research (Allcock et al., 2006).

DNA sequences were compiled and aligned by eye in Se-Al v2.0a11 Carbon (Rambaut 2002). The sequence data for each gene were concatenated into a single data set. Of the 1,612 characters used in the analysis, 177 (11%) were found to be variable.

PAUP v4.0b10 (Swofford, 1998) was used to perform full heuristic searches. The phylogenetic tree is rooted using *Enteroctopus dofleini*, as previous phylogenetic studies using

TABLE 3. Measurements (mm) of ***Benthoctopus rigbyae*** n. sp. Abbreviations: min – minimum; max – maximum; s.d. – standard deviation; c.v. – coefficient of variation; N – number of octopods measured. Measurements were recorded at sea on freshly collected specimens. Total length and dorsal mantle length were recorded when specimens condition was adeqaute whereas other measurements were recorded on a subset of specimens when time permitted. Spermatophore and egg measurements are from the largest of each from N individuals rather than multiple spermatophores or eggs from a single individual.

	min	max	median	mean	s.d.	c.v.	N
Total length	100	400	266	258	68	0.26	78
Dorsal mantle length	18	105	60	60	18	0.31	79
Ventral mantle length	28	85	60	58	15	0.26	27
Mantle width	28	80	65	59	14	0.24	27
Head width	21	49	40	39	7	0.18	28
Eye diameter	11	28	18	19	4	0.20	28
Pallial aperture width	18	50	35	35	7	0.20	27
Full funnel length	15	36	27	27	6	0.21	28
Free funnel length	12	30	19	19	5	0.26	28
Deepest web segment depth	13	55	46	45	9	0.21	24
Arm width (largest arm)	6	20	14	14	3	0.21	28
Left dorsal arm length	128	330	230	219	55	0.25	23
Left dorsolateral arm length	137	285	223	213	45	0.21	22
Left ventrolateral arm length	127	320	223	216	50	0.23	26
Left ventral arm length	124	300	223	217	49	0.23	27
Right dorsal arm length	132	320	215	221	51	0.23	25
Right dorsolateral arm length	130	320	225	220	52	0.24	25
Right ventrolateral arm length	105	280	185	180	43	0.24	26
Right ventral arm length	112	300	221	220	51	0.23	24
Diameter of largest sucker	3	7	5	5	1	0.23	28
Calamus length	4	12	9	8	2	0.25	15
Ligula length	8	26	18	17	5	0.26	15
Spermatophore length	42	104	88	82	23	0.29	6
Spermatophore width	3	3	3	3	0	–	5
Length of largest egg	13	24	18	17	4	0.24	6
Width of largest egg	3	8	7	6	2	0.32	6
Web depth, segment A	8	55	42	40	12	0.31	15
Web depth, segment B	13	55	38	38	13	0.33	15
Web depth, segment C	9	55	44	41	13	0.31	16
Web depth, segment D	11	55	45	42	11	0.26	16
Web depth, segment E	11	50	32	33	11	0.35	15

a wide range of octopodiform species have confirmed that *Enteroctopus* is the sister taxon to *Benthoctopus* (Strugnell et al., 2005) and subsequently that *Enteroctopus* is a suitable outgroup to the genus *Benthoctopus* (Allcock et al., 2006). Starting trees were generated by neighbour joining (NJ) (Saitou & Nei, 1987). A GTR (Γ + I) likelihood model incorporating rate heterogeneity (four rate categories) was used. Branch swapping was performed using TBR (tree-bisection-reconnection). Parameters were then re-estimated and finally branch swapping was performed using NNI (nearest-neighbour interchange). Substitution model parameter values were A = 0.34, C = 0.12, G = 0.15, T = 0.39, $A \leftrightarrow C = 1.48*e^{8}$, $A \leftrightarrow G = 1.53*e^{9}$, $A \leftrightarrow T = 2.80*e^{8}$, $C \leftrightarrow G = 5.55*e^{-11}$, $C \leftrightarrow T = 2.50*e^{9}$ $G \leftrightarrow T = 1.00$, I = 0.81, Γ = 300. ML bootstrap values of

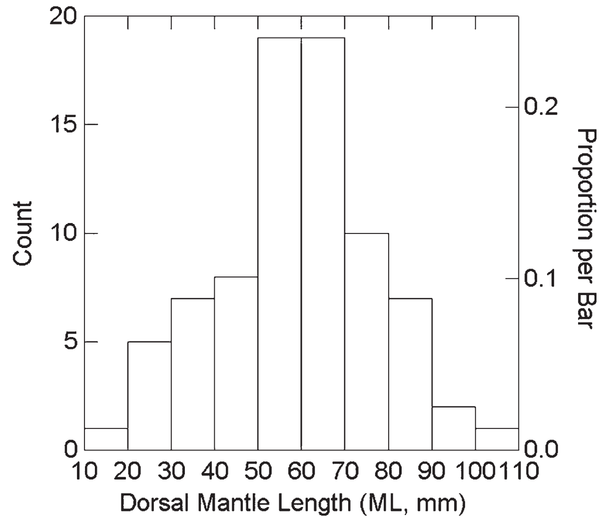

FIG. 1. Size-frequency histogram for 84 specimens of ***Benthoctopus rigbyae*** n. sp. examined in this study.

clade support were generated using the above parameters using 1,000 replicates.

MrBayes v3.1.2 (Ronquist & Huelsenbeck, 2003) was used to calculate marginal posterior probabilities using the GTR + I + Γ model of nucleotide substitution for each partition. Model parameter values were treated as unknown and were estimated in each analysis. Random starting trees were used and analyses were run 1 million generations, sampling the Markov chain every 100 generations. The analysis was performed twice, in each case starting from a different random tree to ensure the analyses were not trapped in local optima. Stationarity was deemed to be reached when the average standard deviation of split frequencies, shown in MrBayes 3.1.2 was less than 0.01 (Ronquist & Huelsenbeck, 2003).

The program Tracer v1.3 (Rambaut & Drummond, 2003) was used to determine the correct "burn-in" for the analysis (i.e., the number of initial generations that must be discarded before stationarity is reached).

RESULTS

The 93 *Benthoctopus* specimens from the Antarctic Peninsula ranged in ML from 18–105 mm. Size-frequency distribution was unimodal with the strong mode from 50–70 mm ML (Fig. 1). The relationship between ML and total length was essentially linear (Fig. 2). Sex ratio was 1.6, male dominant.

Molecular Analyses

Sequences generated in this study are available from GenBank under accession numbers FJ428003–FJ428015.

The sequences of the two ***Benthoctopus rigbyae*** n. sp. individuals from stations 61/048-1 and 42/003 on the Antarctic Peninsula (Fig 3) were identical (PP = 100, BS = 98). These specimens form a monophyletic group with *Benthoctopus* sp. from station 39/014 in the Weddell Sea, which is highly supported by Bayesian posterior probabilities (PP) (PP = 100) and maximum likelihood bootstrap (BS) values (BS = 92). The sequences of *Benthoctopus* sp. from sta 39/014 in the eastern Weddell Sea (Fig. 1) differ from those of ***Benthoctopus rigbyae*** n. sp. by 0.7% (4 base pairs) for the gene 16S rDNA and 2.7% (18 base pairs) for the gene COI.

The sequences of the two *Benthoctopus levis* individuals, captured from Heard Island, were almost identical (differing by only 0.15% (1 base pair) in the gene COI and 0.18% (1 base pair) in the gene 16S rDNA and grouped together in the tree (PP = 100, BS = 100). *Benthoctopus levis* is the sister taxa to *Benthoctopus thielei* (PP = 62, BS = 62) from Kerguelen Island. The clade containing *Benthoctopus levis* and *Benthoctopus thielei* forms a sister taxa relationship to the clade containing ***Benthoctopus rigbyae*** n. sp. and *Benthoctopus* sp. from the eastern Weddell Sea. This relationship is not highly supported however.

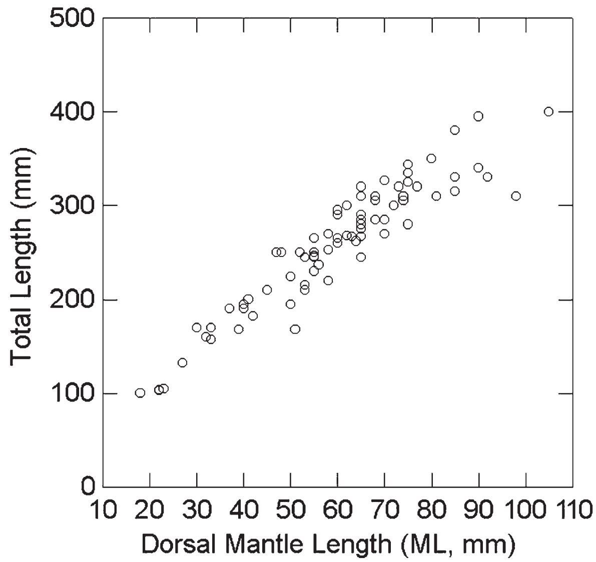

FIG. 2. Relationship between dorsal mantle length and total length of ***Benthoctopus rigbyae*** n. sp.

A sister taxa relationship is present between the clade containing *Benthoctopus* from the Southern Ocean (***Benthoctopus rigbyae*** n. sp., *Benthoctopus* sp. from the eastern Weddell Sea, *B. levis* and *B. thielei*) and *B. johnsoniana*. (PP = 98, BS = 84). *Benthoctopus normani* is basal within the *Benthoctopus* species included in this phylogenetic study.

Systematics

Benthoctopus rigbyae, n. sp.

Holotype: *Polarstern* ANT XIV/2 sta. 42/022, mature male, 64 mm ML, USNM 1117765.

Paratypes: *Polarstern* ANT XIV/2 sta. 42/022, 2 mature females, 55 and 52 mm ML, USNM 1117766.

Other material examined: *Polarstern* ANT XIV/2: Sta. 42/003, 2 immature males, 27 and 48 mm ML, 2 immature females, 24 and 30 mm ML, USNM 1021054; sta. 42/014, 1 immature male, 58 mm ML and 1 mature female, 90 mm ML, USNM 1020987; sta. 42/017, 1 spent? female, 58 mm ML; sta. 42/020, 1 immature male, 39 mm ML; sta. 42/021, 1 mature male, 74 mm ML, 1 immature male, 51 mm ML, 1 juvenile, 18 mm ML; sta. 42/022, 3 immature males, 22–37 mm ML; sta. 42/023, 1 male, mantle and viscera missing, USNM 1021986; sta. 42/029, 2 mature females, 47 and 81 mm ML, 1 male, mantle and viscera missing; sta. 42/036, 2 mature males, 73 and 85 mm ML, 1 mature female, 66 mm ML, 1 immature male, 45 mm ML, 1 immature female, 32 mm ML; sta. 42/040, 5 mature males, 65–75 mm ML, 3 mature females, 48-90 mm ML, 1 immature male, 40 mm ML; sta. 42/041, 1 mature male, 53 mm. ML; sta. 42/043, 1 mature female, 65 mm ML; sta. 42/047, 1 mature female, 50 mm ML; sta. 42/080, 1 mature male, 63 mm ML. *Polarstern* ANT XIX/3: Sta. 61/044-1, 1 mature female, 70 mm ML, NMSZ 2002037.030; sta. 61/045-1, 1 mature male, 98 mm ML, NMSZ 2002037.031; sta. 61/048-1, 1 immature male, 42 mm ML, NMSZ 2002037.032; sta. 61/049-1, 2 immature females, 70-75 mm ML, NMSZ 2002037.033; sta. 61/059-1, 9 mature males, 55-85 mm. ML, 5 immature males, 40–65 mm ML, 2 mature females, each 55 mm ML, 2 immature females, 45-60 mm ML, 1 juvenile female, 30 mm ML, 5 specimens preserved, NMSZ 2002037.034; sta. 61/060-1, 1 immature female, 41 mm. ML; sta. 61/062-1, 1 mature male, 65 mm ML; sta. 61/064-1, 3 mature males, 75–92 mm ML, 1 immature male, 62 mm ML, 1 juvenile male, 33 mm ML, 3 immature females, 56–68 mm ML; sta. 61/101-1, 1 immature male, 60 mm ML; sta. 61/103-1, 2 mature males, 72–85 mm ML, 1 juvenile male, 27 mm ML, 2 mature females,

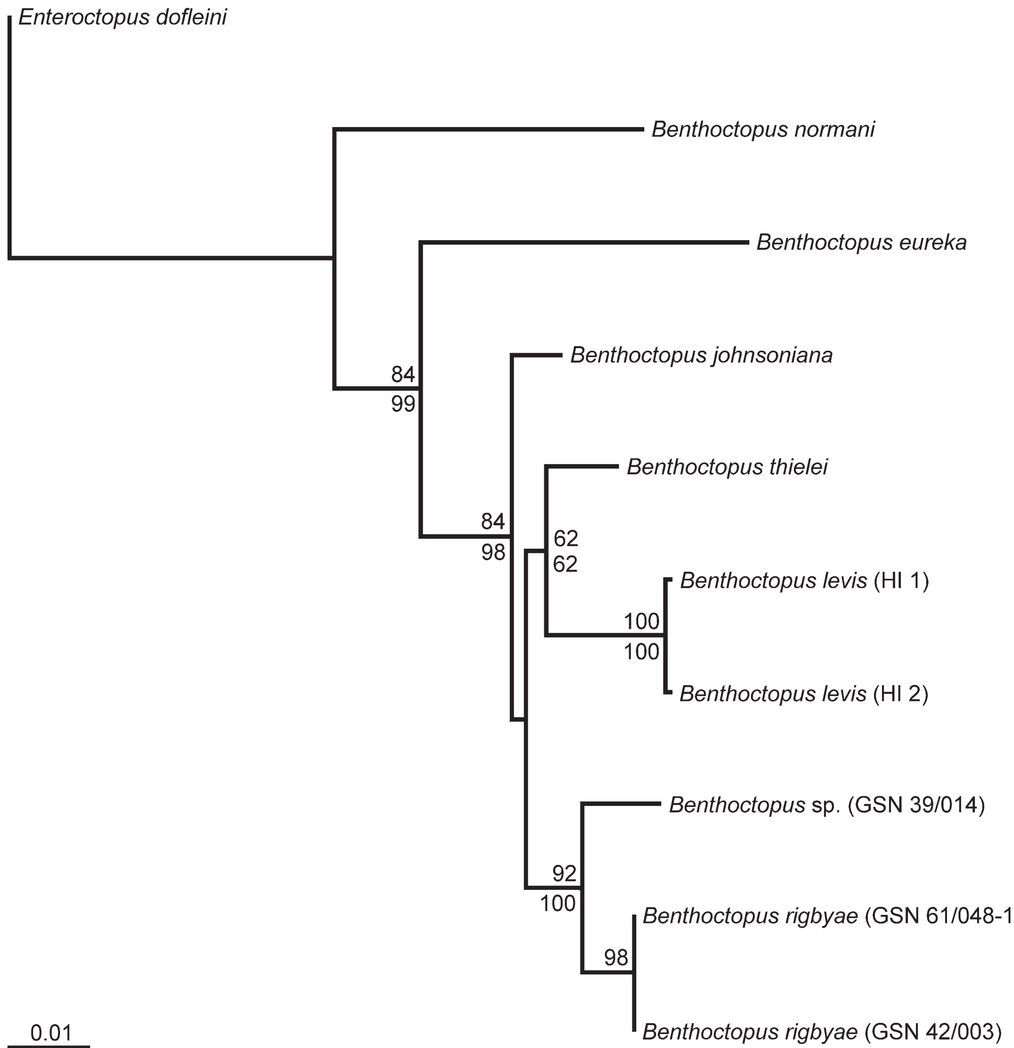

FIG. 3. Maximum likelihood tree depicting the phylogenetic relationship of seven species (ten individuals) of Octopoda. The analysis employed three mitochondrial sequences (12S rDNA, 16S rDNA, COI) concatenated. Bayesian support values are indicated below the nodes, maximum likelihood bootstrap values are indicated above the nodes.

80–105 mm ML, 1 immature female, 60 mm ML; sta. 61/107-1, 1 juvenile male, 23 mm ML; sta. 61/109-1, 1 mature female, 53 mm ML, 1 immature female, 53 mm ML; sta. 61/111-1, 1 juvenile male, 22 mm ML. *Polarstern* ANT XXIII/8: sta. 69/609-1, 1 juvenile, 24 mm ML; sta. 69/616-1, 1 immature female, 41 mm ML; sta. 69/652-1, 1 immature female, 20 mm ML; sta. 69/654-6, 1 immature female, 32 mm ML; sta. 69/661-2, 1 immature female, 57 mm ML; sta. 69/662-1, 1 mature male, 61 mm ML; sta. 69/663-1, 1 immature female, 47 mm ML, sta. 69/664-1, 2 immature female, 34–49 mm ML.

Diagnosis

Antarctic octopod with biserial suckers and W-shaped funnel organ, lacking an ink sac. Arms long, 72–94% of total length, with shallow webs approximately 20% of longest arm

TABLE 4. Counts from specimens of ***Benthoctopus rigbyae*** n. sp. See Table 3 for abbreviations.

	min	median	mean	s.d.	c.v.	max	N
Suckers on hectocotylized arm	61	66	67	4	0.06	78	15
Suckers on left ventrolateral arm	113	148	144	14	0.1	165	25
Suckers on longest arm	124	146	144	11	0.08	164	24
Inner gill lamellae	5	7	7	0.7	0.12	8	27
Outer gill lamellae	6	6	7	0.6	0.1	8	27

length. Hectocotylized arm with 60–80 suckers, 56–75% length of longest arm. Ligula length 6–16% of hectocotylized arm length.

Description

Measurement, counts, and indices and their associated statistics are summarized in Tables 3–5.

General Appearance of Live/Fresh Animal (Fig. 4): A moderately large octopod, to at least 105 mm ML, 400 mm TL. Chromatophores very tiny. Overall color light brownish orange, grading to lighter color ventrally and orally but darker at bases of suckers on some animals. Some animals showed a blotchy pattern on the mantle and web, but no changes in color pattern were noted during observation of live animals. Skin smooth, without papillae or superocular cirri. Mantle with obvious dorsal hump, no distinct lateral ridge or keel, although some animals exhibited a band of raised skin laterally on mantle. Arms long. Web formula variable; web shallow, extends along ventral arm to tip and along dorsal arm, attaching to aboral arm surface approximately 1/3–1/2 arm length from proximal. Eyes prominent, dark, with bluish sheen. Mantle muscular.

External Morphology (Fig. 5): Head narrower (approximately 67%) than mantle, head width about 63% ML. Mantle width only slightly less than ML. Arms often regenerating but very long,

TABLE 5. Indices from specimens of ***Benthoctopus rigbyae*** n. sp. See Table 3 for abbreviations.

	min	median	mean	s.d.	c.v.	max	N
Dorsal mantle length / Total length	0.18	0.23	0.23	0.03	0.11	0.32	78
Ventral mantle length / Dorsal mantle length	0.77	0.91	0.91	0.08	0.08	1.05	27
Mantle width / Dorsal mantle length	0.72	0.93	0.93	0.09	0.09	1.15	27
Head width / Dorsal mantle length	0.46	0.61	0.62	0.08	0.13	0.79	27
Head width / Mantle width	0.53	0.64	0.67	0.08	0.12	0.84	27
Eye diameter / Dorsal mantle length	0.22	0.29	0.30	0.04	0.14	0.38	27
Full funnel length / Dorsal mantle length	0.36	0.44	0.43	0.05	0.12	0.54	27
Free funnel length / Dorsal mantle length	0.21	0.31	0.30	0.05	0.15	0.41	27
Deepest web depth / Dorsal mantle length	0.41	0.71	0.72	0.15	0.20	0.98	23
Deepest web depth / Longest arm length	0.09	0.18	0.19	0.03	0.18	0.25	24
Calamus length / Dorsal mantle length	0.09	0.12	0.13	0.02	0.14	0.15	14
Calamus length / Hectocotylized arm length	0.03	0.05	0.05	0.01	0.24	0.08	15
Ligula length / Dorsal mantle length	0.18	0.27	0.26	0.03	0.13	0.30	14
Ligula length / Hectocotylized arm length	0.06	0.10	0.11	0.03	0.25	0.16	15
Longest arm length / Dorsal mantle length	2.50	3.77	3.75	0.63	0.17	4.89	27
Longest arm length / Total length	0.72	0.83	0.83	0.07	0.08	0.94	25
Hectocotylized arm length/Opposite arm length	0.56	0.76	0.74	0.09	0.12	0.90	15

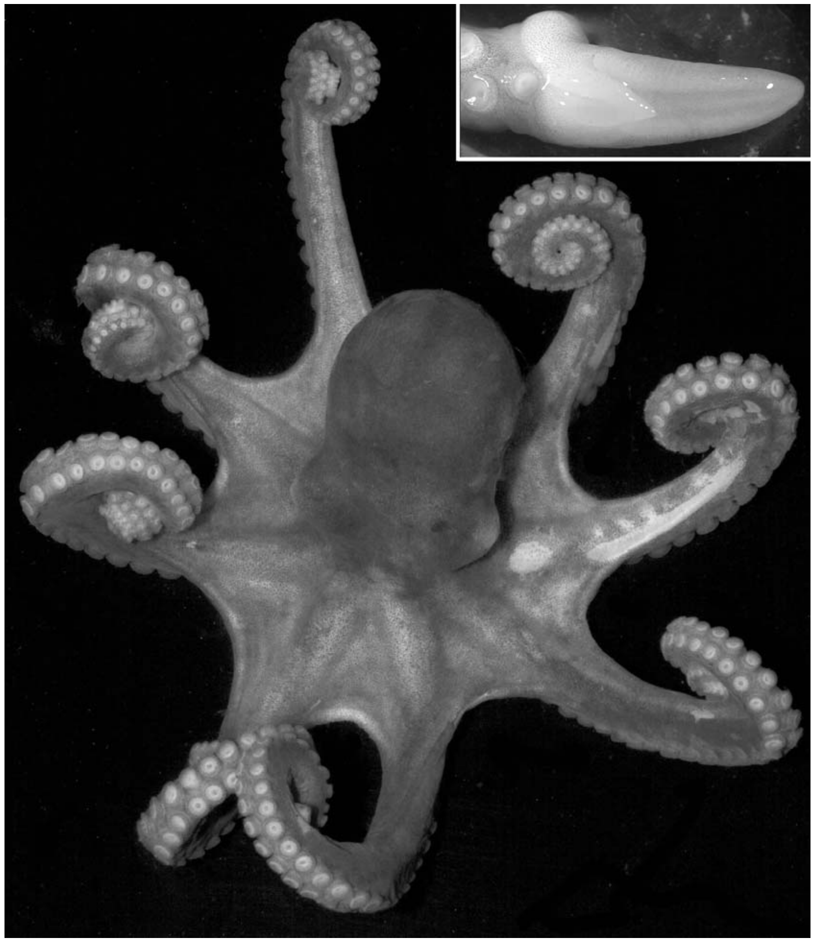

FIG. 4. Shipboard photograph of a live animal of ***Benthoctopus rigbyae*** n. sp. (by E. Jorgensen, 2007). Note nearly uniform color, lack of papillae and lack of skin components. Inset: Close-up of the hectocotylized arm tip of a mature male.

comprising 70–90% of total length, generally 3–4 times ML. Order of arm length variable but non-hectocotylized arms typically subequal in length, with 113–165 suckers. Variability in sucker counts partly a result of difficulty in counting very many pairs of miniscule suckers on arm tips. Hectocotylized right ventrolateral arm (Fig. 6a) length approximately 75% of opposite arm length. Sucker number on hectocotylized arm 61–78. Suckers small, maximum diameter 1–3% of arm length or approximately 7.5% ML. Sucker series widely separated, with zigzag pattern of transverse ridges and grooves between adjacent suckers (Fig. 6a). Distinctly enlarged suckers absent. Ligula with indistinct median ridge and numerous faint transverse creases that sometimes are barely visible. Ligula length approximately 10% (6–16%) of hectocotylized arm length or 26% (18–30%) of ML. Calamus sharply pointed, length approximately half (0.46–0.50%) of ligula length. Funnel length approximatrely 1/3–1/2 ML; length of free funnel approximately 20–40% ML.

Internal Anatomy: Funnel organ W-shaped, with moderately thick limbs. Chromatophores present in lining of buccal cavity. Beaks as in Figure 6e–g. Radula (Fig. 6j): rachidian multicuspid, 2 pairs laterals small and triangular, 2nd larger than 1st, 1 pair marginal teeth curved

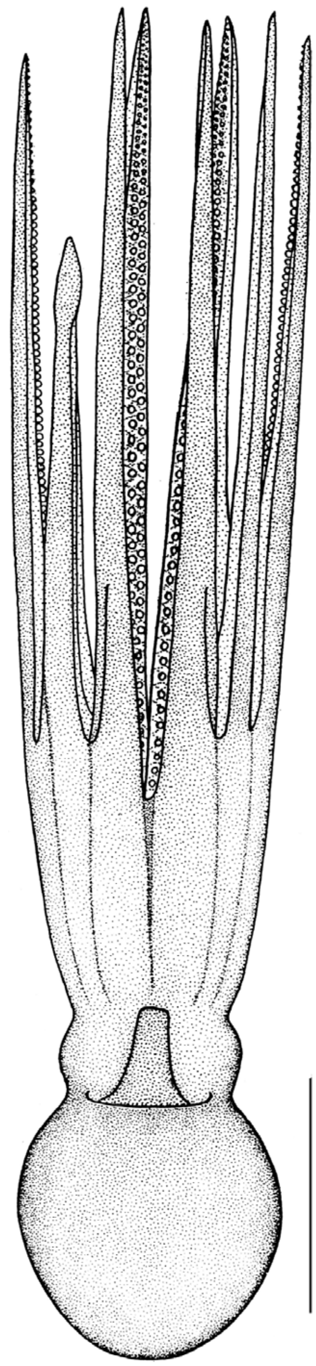

FIG. 5. ***Benthoctopus rigbyae*** n. sp. external anatomy, ventral view, male, 70 mm ML. Scale bar = 50 mm.

and much larger than laterals, 1 pair marginal plates. Dorsal visceral membrane densely covered with chromatophores. Digestive gland (Fig. 6h, i) almost spherical but slightly flattened dorso-ventrally, with shallow depression for esophagus, stomach and caecum, cream to deep maroon in color. Esophagus straight, slightly expanded in crop region, no crop diverticulum. One octopod when dissected had ventral area of crop region swollen and spherical with fluid. Anterior salivary glands large, dorsolateral on buccal mass. Posterior salivary glands moderately large, each similar in size to stomach. Stomach and caecum about equal in size. Intestine straight. Male reproductive tract as illustrated in Figure 6c, d. Accessory spermatophore gland longer than Needham's sac. An 80 mm ML male had 28 mature spermatophores in Needham's sac; one of 63 mm ML had 24, with lengths ranging from 55 to 102 mm (mean 94 mm). Female reproductive tract as illustrated in Figure 6b. Oviducts narrow, exit from antero-ventral membrane of ovary separately but contiguously, then bend laterally and funnel into round oviducal glands. In mature females, distal oviducts longer than proximal, white and thick, diameter only slightly less than that of oviducal gland. Mature ovarian eggs elongate, 13–24 mm long (mean 17 mm) x 3–8 mm wide (mean 6 mm). An 81 mm. ML female had 88 mature and 4 immature (3 x 8 to 3 x 10 mm) eggs in ovary.

Size at Maturity: Maturity of males increases gradually with increasing size (Fig. 7). Females, on the other hand, exhibit a wide range of maturity stages over a fairly narrow size range (Fig. 8). This may indicate that although most females appear to attain larger sizes than males, females mature rapidly once a threshold size has been reached.

Stomach Contents: Of 15 stomachs opened (n = 9 females, 6 males), eight were empty (n = 6 females, 2 males) and seven were partly filled but not distended (n = 3 females, 4 males). Two stomachs contained only amphipod remains; one contained amphipod and fish remains in approximately equal volumes; one contained half amphipod remains together with the remains of serolid isopods, polychaetes, bryozoans and sponge spicules. One stomach contained solely the remains of ophuroids. Two stomachs contained unidentifiable crustacean remains.

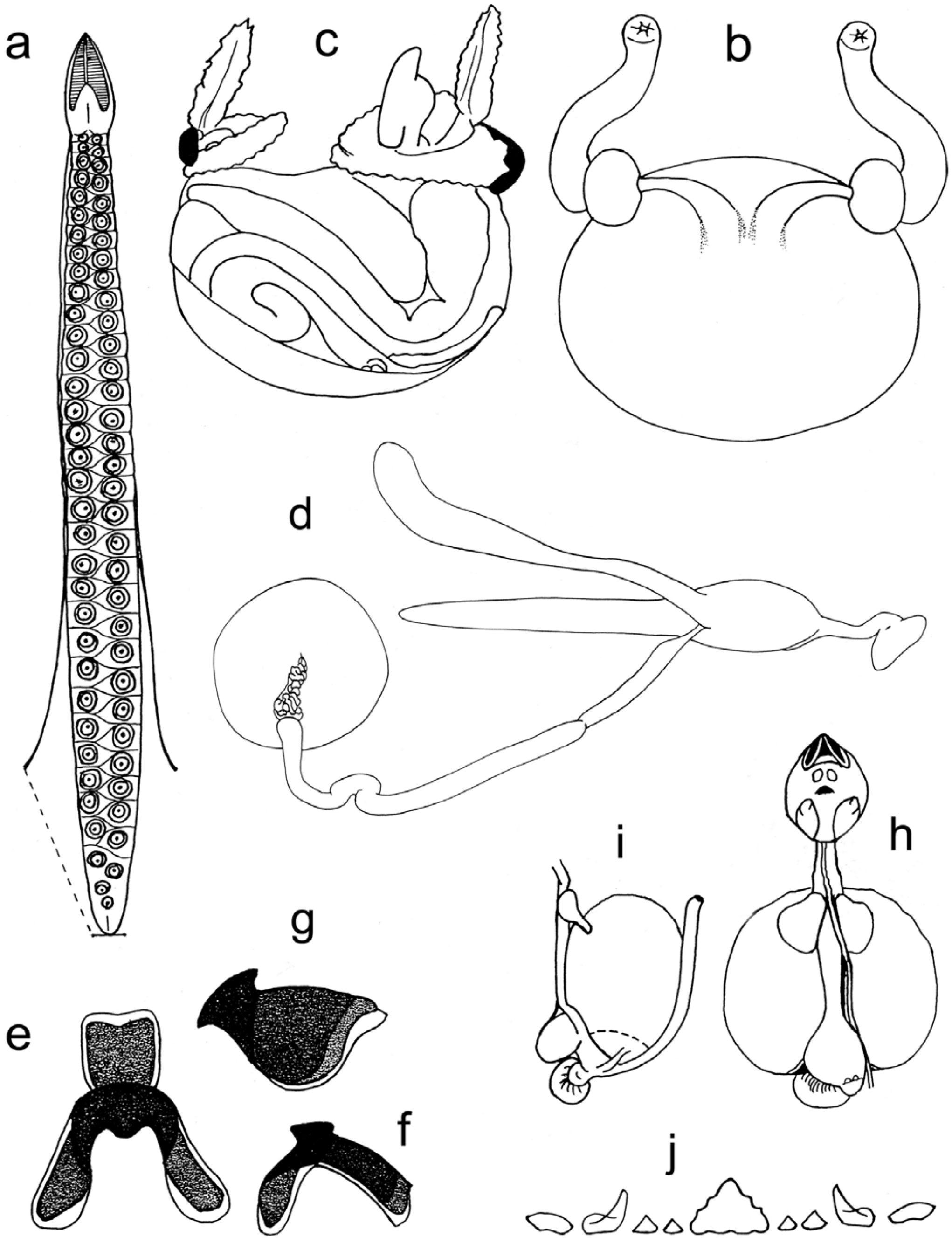

FIG. 6. ***Benthoctopus rigbyae*** n. sp. Internal anatomy (a) hectocotylized arm, (b) female reproductive system, (c) male reproductive system as seen in the viscera, (d) male reproductive system dissected to show structure, (e) lower beak, ventral view, (f) lower beak, lateral view, (g) upper beak, lateral view, (h) digestive system, dorsal view, (i) digestive system, lateral view, (j) one row of radular teeth.

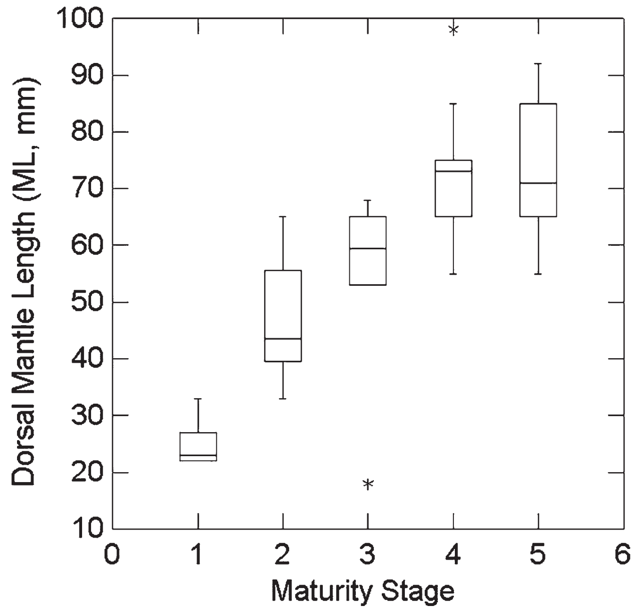

FIG. 7. Relationship between maturity stage and dorsal mantle length for male ***Benthoctopus rigbyae*** n. sp.

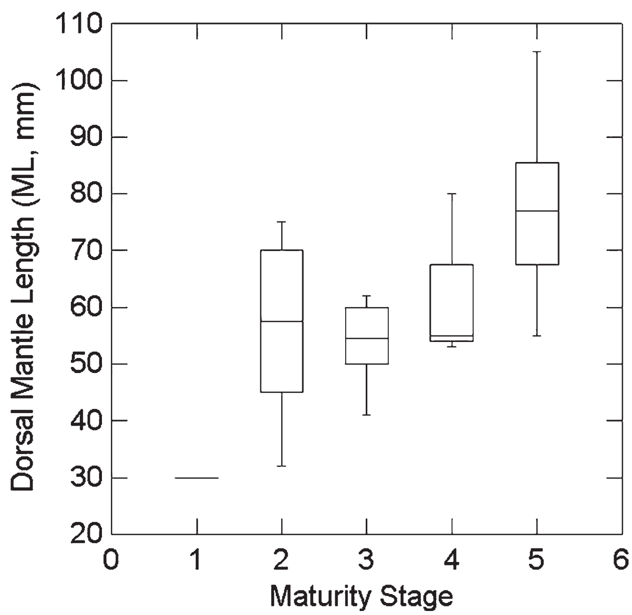

FIG. 8. Relationship between maturity stage and dorsal mantle length for female ***Benthoctopus rigbyae*** n. sp.

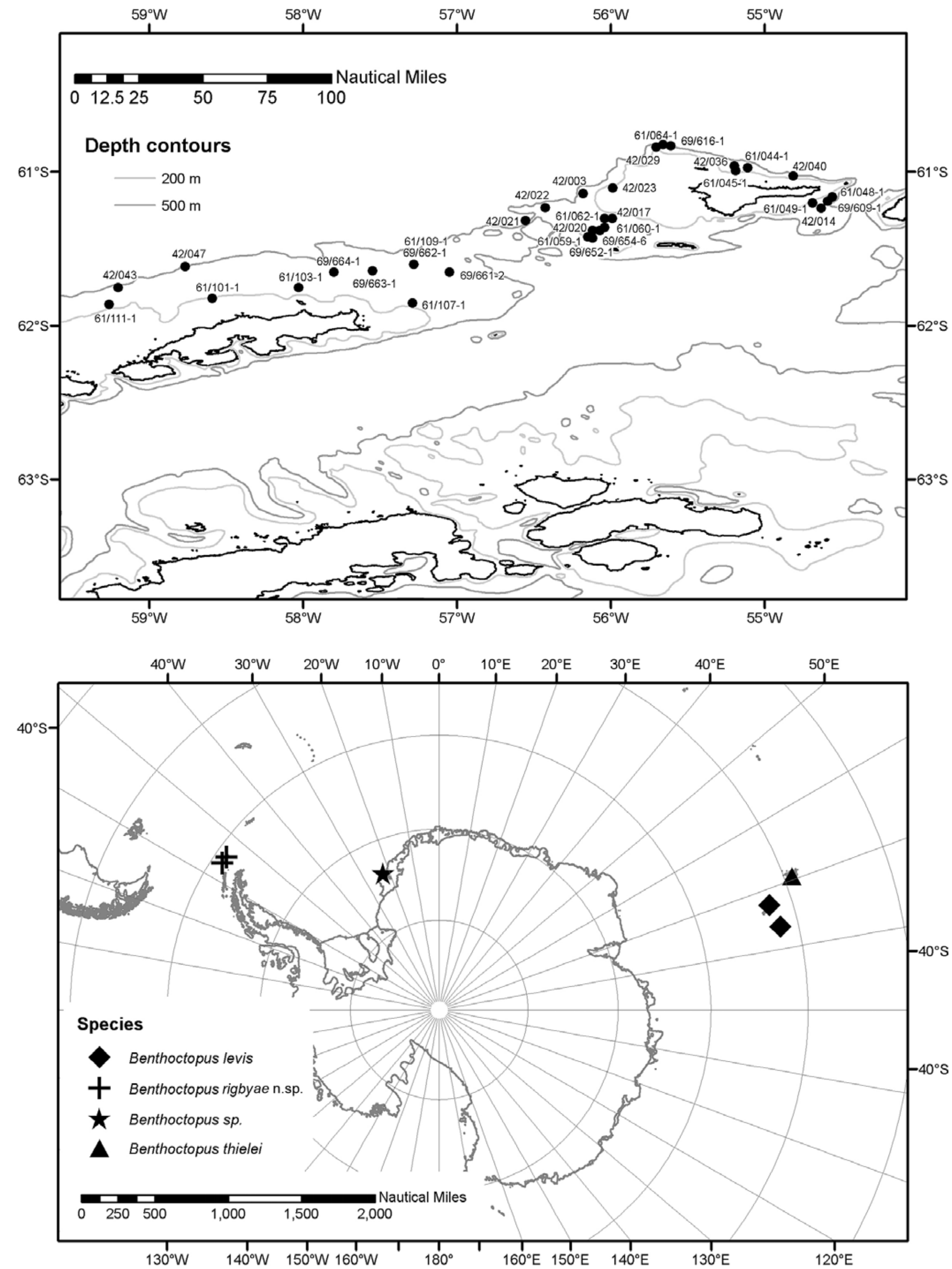

FIG. 9. Distribution of ***Benthoctopus rigbyae*** n. sp. catches in the vicinity of the South Shetland Islands (top); Distribution of *Benthoctopus* spp. around Antarctica (bottom).

Length/Weight Relationship: The relationship between total length and weight, including all maturity stages, was similar between the sexes. For females the relationship could be described by the power function $y = 0.0492x^{1.9845}$, $r^2 = 0.8984$. The function descriptive of the males is $y = 0.0023x^{2.6887}$, $r^2 = 0.9298$

Distribution

These octopods were common, though not abundant, along the outer shelf and upper slope of entire South Shetland Island chain, depth range 250–600 m (Fig. 9). They comprised < 2% of the octopod fauna sampled. The maximum number collected in a 30-min tow with the bottom trawl was 18.

DISCUSSION

Octopods with biserial suckers but lacking an ink sac currently are considered to belong to either *Bathypolypus* or *Benthoctopus.* The former is fairly well defined (Muus, 2002), based primarily on morphology of the hectocotylus, which has a very large ligula characterized by prominent transverse laminae. Although the males described here have fairly large ligulae with very faint transverse creases, their long arms, small suckers, and smooth skin make them more similar to *Benthoctopus* than *Bathypolypus. Benthoctopus,* however, has tended to be a "catch-all" taxon including any such octopod that does not clearly belong to *Bathypolypus.* The genus as currently defined probably is not holophyletic. Species of the genus *Benthoctopus* described from the area of the Antarctic Polar Frontal Zone (APFZ) or further south include just *B. theilei* Robson, 1932, and *B. levis* (Hoyle 1885). The type localities of *B. theilei* and *B. levis* lie on the APFZ off the Kerguelen Islands and south of the APFZ off Heard Island respectively.

Based on published descriptions, the specimens described here are most similar to *B. levis*. However, they differ in relative arm length, web depth, lengths of the hectocotylized arm and ligula. One of us (L.A.) examined the holotype of *B. levis* (BMNH 1889.4.24.43) and found that its funnel organ is VV-shaped rather than W-shaped. A photograph of *B. levis* from near the type locality published by Norman (2000) shows an octopod similar to ours, but with shorter arms and a deeper web. ***Benthoctopus rigbyae*** is easily distinguished from *B. thielei* as the latter has a VV funnel organ (illustrated by Robson, 1932: 234).

Although similar to *B. levis,* the specimens we describe differ sufficiently in morphology for us to believe they represent a separate species. This conclusion is supported by the molecular data. ***Benthoctopus rigbyae*** n. sp. is the sister taxon to *Benthoctopus* sp. from the eastern Weddell Sea, whilst *B. levis* is the sister taxon to *B. thielei*. The Weddell Sea specimen differs from ***B. rigbyae*** by 18 base pairs in COI the DNA bar-coding gene. We must therefore conclude that the Weddell Sea specimen also represents a different, currently undescribed species. Measurements taken shortly after capture support this. However, this specimen was destroyed due to mechanical failure of a freezer prior to fixation and is no longer extant.

To discover two species in the western Southern Ocean is not surprising given that these *Benthoctopus* species appear to inhabit shallower waters than they do elsewhere. ***Benthoctopus rigbyae*** is known from 200–600 m and the Weddell Sea specimen was captured at 850 m. Between the South Shetland Islands and the eastern Weddell Sea, water depths exceed the maximum known depth for the species. Given that mature ovarian eggs of ***B. rigbyae*** have been recorded at 24 mm length and undoubtedly hatch into benthic crawl-away young, opportunities for gene flow between the Peninsula region and the eastern Weddell Sea would be extremely limited. Thus, there is limited possibility of a continuous population around the Antarctic continent because of the Filchner-Ronne Ice Shelf which extends out over water depths greater than 1,000 m.

It is perhaps more surprising to find clearly differentiated species between the Kerguelen Islands and Heard Island. The geographic distance between these islands is small and both are situated on the Kerguelen plateau meaning that there is no deep water trench between them. It is possible that the APFZ in this region is influential in delimiting two distinct microhabitats.

Etymology

Named in memory of Robin Rigby, a bright, young cephalopod biologist.

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